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BIOCHEMICAL VARIABILITY DURING SATURATION DIVING TRAINING.(U)
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INTRODUCTION

Recent research has shown that serum levels of cholesterol and uric acid, which had been previously regarded as relatively stable, are in fact quite variable. Variation in levels of serum cholesterol and uric acid occurs both prior to and during situations that may be broadly classified as stressful. Elevations of serum cholesterol have been noted in students prior to an examination (1) and prior to stressful cold exposure (2). However, men facing job loss after many years of continuous employment did not show elevated serum cholesterol levels prior to expected discharge but did have elevated serum cholesterol levels after being unemployed (3).

Elevations of serum cholesterol occurring during stressful situations have been related to a number of situational, emotional and personal factors. A situational factor common to many reports (4) is work overload. Objective work overload results in serum cholesterol elevation. However, subjects who performed low work loads but subjectively experienced high work loads had lower cholesterol levels than subjects who both performed and experienced low work loads. The experience of succeeding on a task thought to be difficult lowered serum cholesterol. Any judgment about the relative difficulty of a task is based in part on the subject's previous experience on similar tasks and suggests that serum cholesterol responses in stressful situations should be related to previous stressful experience.

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The role of prior experience in stress-induced serum uric acid elevations is somewhat more evident. Rahe noted the UDT trainees showed serum uric acid elevations prior to novel situations, but none in situations involving strenuous practice of techniques previously learned (5). In another study, serum uric acid rose in anticipation of job loss after many years of continuous employment (4).

Although stress-induced serum uric acid elevations have been linked to motivation and leadership (6), it is possible that situational elements, such as novelty, may also play an important role.

The U.S. Navy saturation diving training course consists of training periods during which stresses of varying nature and severity are imposed. In addition, the trainees, though they were all first class divers, had varying experience with the techniques and concepts taught in the training course. In this longitudinal study, it was expected and found that elevations of serum cholesterol and uric acid occurred during stressful portions of the course and that such elevations were also related to the diver trainees' previous experience.

METHODS

Subjects

The 31 men chosen for the saturation diving training course were U.S. Navy first class divers. The mean age of the trainees was 28 years, \pm 4 years. Each man was given a thorough medical examination prior to the start of training. Three men were eliminated from the training program for medical reasons. There was no history of metabolic, cardiovascular disease, or of gout in any of the subjects continuing in the training program. Levels of physical fitness varied among the trainees. To prepare all the men for the physical requirements of

saturation diving, a physical training schedule was initiated and continued throughout the training period. This consisted of 20 minutes of calisthenics and a 2- to 4-mile run daily and a 1500-yard swim twice weekly. By the end of the first training week, all trainees met the physical fitness standards required by the medical department for saturation diving.

The average educational level of the trainees was 11.6 years, \pm 1.5 years. As first class divers, all the men had had formal training and some experience with all common types of diving equipment, including deep sea helium-oxygen apparatus. However, none had had experience with saturation diving equipment. Actual diving experience was ascertained by a Diving Experience Questionnaire administered prior to training. This questionnaire assessed the number of dives, their depths and the equipment used as well as the incidence of diving accidents such as bites, stings, and the various forms of decompression sickness. At the same time, a 40-item mood check list and diet and activity form was completed and a control blood sample obtained.

The saturation diving training course is a 14-week program consisting of classroom instruction, training and supervised experience with aquanaut equipment, and training and experience in the use of a complete saturation diving system. Due to equipment limitations and because much of the instruction was individualized, only a limited number of students (16) could be accommodated during some of the training periods. Trainees in two successive courses were studied. The course content and the characteristics of the trainees were essentially identical during both 14-week sessions. The first week of the course consisted of an intensive review of the biology and the physics of diving followed by a written examination

on the material covered. Although the trainees knew there was little probability that anyone would be eliminated from the program for academic reasons, the emphasis placed upon the examination and the trainees' limited academic background and their inexperience in classroom study caused considerable stress and some embarrassment. In some cases, the trainees requested and received individual tutoring. On the day before the examination, and the day after, the mood and activity form was administered and a blood sample obtained from each trainee.

The next several weeks of the training program consisted of progressive training in the use of aquanaut equipment under various conditions. The principal item introduced at this time was the Mark 8 UBA, a moderately complex diving rig worn on the diver's back, composed of inhalation and exhalation bags, a CO₂ absorption canister and emergency breathing gas. The diver breathes ^ahelium-oxygen mixture supplied by a surface-connected umbilical. Also introduced at this time were the Mark 11 UEA, the KMB-9 face mask and the hot water wet suit. The training sequence progressed rapidly, beginning with use of the equipment in a swimming pool, then to shallow pier dives, and finally to 180 feet open sea dives. A blood sample was obtained and a mood and exercise form was completed by all trainees prior to the pier dive.

The saturation dive was the most unique event the trainees faced in the program as none of them had had any prior experience with this diving technique. In non-saturation diving, the amount of inert gas absorbed by the tissues and which must be safely released before the diver can surface, is determined by the depth and duration of the dive. In a saturation dive a maximum amount of inert gas is absorbed by the tissues after 12 hours' exposure at a given depth. Thus, decompression from a saturation dive is determined only by the depth of the dive,

provided the duration at that depth exceeds 12 hours (7). The saturation diving system used in this program consisted of a shipboard decompression chamber, a personnel transfer capsule (PTC), and associated life support equipment. The system maintained a constant hyperbaric environment yet allowed the divers to enter the water to work via the PTC.

In this training situation, four trainees and two instructors entered the chamber the evening prior to the dive and were compressed to 180 FSW (95 PSI) in an atmosphere of 95.5% helium, 4.5% O₂, and 0.5% CO₂. The following morning, after a complete equipment check, two of the trainees and ^{one} ~~the~~ instructor entered the PTC and were lowered into the water to 180 feet. Dressed in aquanaut equipment, the trainees entered the water for a period of 30-90 minutes and performed a number of tasks and maneuvers while swimming at 180 to 230 feet. After completing their tasks, the trainees returned to the PTC which was raised and reattached to the chamber. Their places were taken by the remaining two trainees and the sequence repeated. When the in-water phase of the dive had been completed, the trainees reentered the chamber, decompression was begun at 2.5 feet/hour and lasted for 72 hours. Minor decompression sickness was treated with 100% oxygen by face mask. During this period the trainees were in an environment of constant temperature (85°F) and humidity and occupied themselves by watching movies and playing cards. The trainees not involved in the in-water phase of training stood watches and maintained the life support equipment of the chamber and capsule, as part of the training program. The training was completed when a trainee had made a saturation dive and stood support watches on three other saturation dives. The saturation dive provided several unique situations for the trainees including

(a) the use of aquanaut equipment while diving from a capsule, (b) the experience of long-term pressurization and prolonged decompression, (c) the experience of prolonged helium-oxygen exposure with its attendant problems of communication and temperature control, (d) the experience of living in a small space with five other people and minimal privacy, and (e) the knowledge that in the (in-water) event of an emergency, survival was possible only by return to the capsule and that surfacing rapidly would be instantaneously fatal. These features combined to make the saturation dive a unique psychologic and physiologic experience. A blood sample was obtained and mood and activity form completed by each trainee prior to entering the chamber before the dive, at the start of decompression, and at the termination of decompression.

The diving medical officer and two experienced instructors rated each course training period in terms of the stress imposed on each trainee. A scale of 1 (least stressful) to 10 (most stressful) was used in making this evaluation. All blood samples obtained during the study were immediately centrifuged and the serum collected and frozen at -20°C until analyzed. Total serum cholesterol was determined by the method of Clark (8). Serum uric acid was determined by a modification of the enzymatic method of Liddle (9) in which the serum proteins were precipitated with trichloroacetic acid after the enzymatic conversion of uric acid to allantoin. This modification reduced the variance, especially when analyzing lipemic samples.

Serum uric acid and cholesterol values were examined by an analysis of variance procedure for repeated measures ~~ANCOVA~~. Specific differences between the mean values for the various periods in the training program were tested by the Newman

Keuls multiple range test (11). The relationships between items or groups of items on the diving history questionnaire, and serum cholesterol and uric acid values, were determined by standard correlation and partial correlation techniques (12).

RESULTS

Figure 1 illustrates the mean levels of serum cholesterol and uric acid observed during the seven training periods studied. The figure also illustrates the levels of stress imposed on the trainees during each of the training periods. The most stressful periods of the course were the pre-examination period and the periods before and during the saturation dive. Serum cholesterol levels rose from the control period to the pre-examination period and then fell markedly and significantly after the examination. They reached their lowest levels during the pier dive. A significant rise in serum cholesterol levels occurred prior to the saturation dive and the levels remained elevated after decompression. Uric acid levels remained relatively constant throughout the course except during decompression when a marked and significant decline was noted. Uric acid levels had not returned to pre-dive levels at the end of decompression.

Table 1 depicts the relationships between serum uric acid and cholesterol levels and the various facets of diving experience during the three most stressful periods of the course. Age was positively correlated with serum cholesterol levels during all three stressful periods, but the relationship was statistically significant only during the periods before and during the saturation dive. Educational level was apparently unrelated to cholesterol levels during the three stressful training periods. The number of years spent as a diver was related

to serum cholesterol levels before the examination. The higher cholesterol levels of men who had spent many years diving is attributable to their greater age as is indicated by the partial correlation of years diving with cholesterol. The number of dives to depths between 200 and 300 feet was directly and significantly correlated with serum cholesterol levels before and during the saturation dive but was not significantly correlated with the serum cholesterol levels noted prior to the examination. The number of dives made to shallower depths (down to 150 feet) and the number of dives made using hard hat equipment and breathing air was not significantly related to the divers' serum cholesterol levels before or during the saturation dive.

The relationship between the number of dives to depths between 200 and 300 feet and serum cholesterol levels is only partly attributable to the effect of age on serum cholesterol. For the saturation dive period, removal of the effect of age by partial correlation does not appreciably alter the relationship between the number of deep dives and the divers' serum cholesterol levels. However, for the period prior to the saturation dive, removal of the effect of age by partial correlation did render statistically insignificant the correlation between the number of deep dives and serum cholesterol levels.

The only item of the Diving History Questionnaire that was significantly related to serum cholesterol levels during the period prior to the exam was the number of dives made by using hard hat diving gear and breathing a helium-oxygen gas mixture. This item was also correlated with the serum cholesterol levels observed during the control period prior to training. Removal of the effect of age by partial correlation did not appreciably alter the relationship.

A factor analysis of the responses to the Diving History Questionnaire by 180 naval divers in the fleet produced a four-item factor (number of times the bends occurred, number of dives made using semi-closed circuit SCUBA, number of times the respondent served as an experimental subject, and the number of times the respondent tested diving equipment) which was called the Exotic Diving Experience factor. This factor was inversely correlated with the serum uric acid levels observed before and during the saturation dive. The negative correlation indicated that trainees who had had prior exotic diving experience had lower serum uric acid levels before and during the saturation dive. Prior exotic diving experience was not significantly related to the serum uric acid levels before the examination.

DISCUSSION

A number of recent studies have shown that different stressful situations induce changes in serum cholesterol and uric acid levels. Serum cholesterol levels have been shown to rise prior to examinations (1), prior to stressful interviews ¹³ (17), and before cold exposure (2). Elevations of serum cholesterol can also occur during a stressful situation. Elevations of serum cholesterol have been noted during stressful UDT training (5) and during experimentally imposed work overload (4).

During the study of saturation diving training, elevations of serum cholesterol were noted during two distinctly different stressful situations. Such increases occurred prior to the written examination and prior to, during, and after the saturation dive. Cholesterol levels rose before the examination and fell markedly afterward. They rose again prior to the saturation dive and remained elevated thereafter. The elevations of serum cholesterol corresponded to the most stressful periods of the training course but were not strictly

proportional to the stress levels estimated by the instructors and the medical officer. The lack of proportionality suggests that the objective stress of the situations was not the only factor involved in the observed increases in serum cholesterol, although the latter was of major importance. Several studies of students have shown increases of serum cholesterol prior to examinations (1). Sales has shown that objective work overload is sufficient to elevate serum cholesterol levels acutely (4). In this study, similar stress levels induced similar serum cholesterol responses even though the stressful situations were quite different.

Although the instructors placed great emphasis on the examination, the trainees knew there was little likelihood of anyone being eliminated from the training program because of poor academic performance. The question arises as to why the examination was so stressful as to induce serum cholesterol alterations. It seems unlikely that the stress was related to the act of taking the examination or to the testing of the trainee's general knowledge. Educational level was not significantly related to the cholesterol response. However, those trainees who had had previous experience with helium-oxygen diving equipment, and who presumably were more familiar with exotic equipment and procedures, had lower cholesterol levels prior to the examination. This suggests that all the trainees were well aware of the extent of the material about which they were to be tested, and that those who, by reason of prior experience had greater familiarity with the material, felt less anxious and more confident of their ability to perform well on the examination. Medical students who expected to perform poorly had higher serum cholesterol levels (5). Thus, it would seem that the trainee's assessment of the expected stressful situation, and his assessment of his ability

to perform therein contribute to his serum cholesterol response. The serum cholesterol responses during the saturation dive support this generalization. Those trainees who had made more deep dives, and who would thus be expected to be able to estimate accurately the problems and difficulties that might be encountered in the saturation dive, had higher cholesterol levels during the dive. Since none of the trainees had previous saturation diving experience, no estimate can be made of their conception of their ability to perform during the dive. Thus, it would seem that whatever the physiologic mechanisms controlling serum cholesterol, the response is dependent on relatively specific assessments of the situation based on specific experience.

During saturation diving training serum uric acid levels remained relatively constant until decompression from the saturation dive, when a marked and significant drop occurred. There was no change in serum uric acid level prior to the examination. Other studies have also shown that serum uric acid does not rise prior to examinations.^(12,13) No item or groups of items from the Diving History Questionnaire were found to correlate significantly with serum uric acid levels prior to the examination. However, before and during the saturation dive, a group of four interrelated items involving exotic diving experience was found to be negatively related to serum uric acid levels. The four items together related to a class of experience rather than to a specific type of experience. The exotic experience items are similar to the experiences the trainees could undergo during the saturation dive and decompression. The inverse correlation between exotic experience and serum uric acid levels suggests that the serum uric acid response in a stressful situation is related to the relative novelty of the situation.

This is in contrast to the serum cholesterol response which seems to be related to specific assessments of the situation based on relevant experience.

The relationship of serum uric acid response to novelty situations does not contradict the postulated relationship of serum uric acid to motivation (6). It is likely that highly motivated persons would be more likely to volunteer for exotic diving projects such as the testing of equipment. However, if motivation were the sole factor to be considered in this study, then highly motivated individuals who had previously volunteered for exotic diving projects should have had higher serum uric acid levels during the stressful portions of training. It is possible that serum uric acid levels of such individuals might have been higher during the saturation dive if they had not had the previous exotic diving experience. The complex relationships of motivation, experience, and serum uric acid levels during stress are currently under investigation.

The decline in serum uric acid levels during decompression which has not been previously described in the literature, although related to previous diving experience, is probably not caused by experiential or motivational factors. Research currently in progress indicates that such decline during decompression is possibly related to decompression-induced changes in cell membrane permeability.

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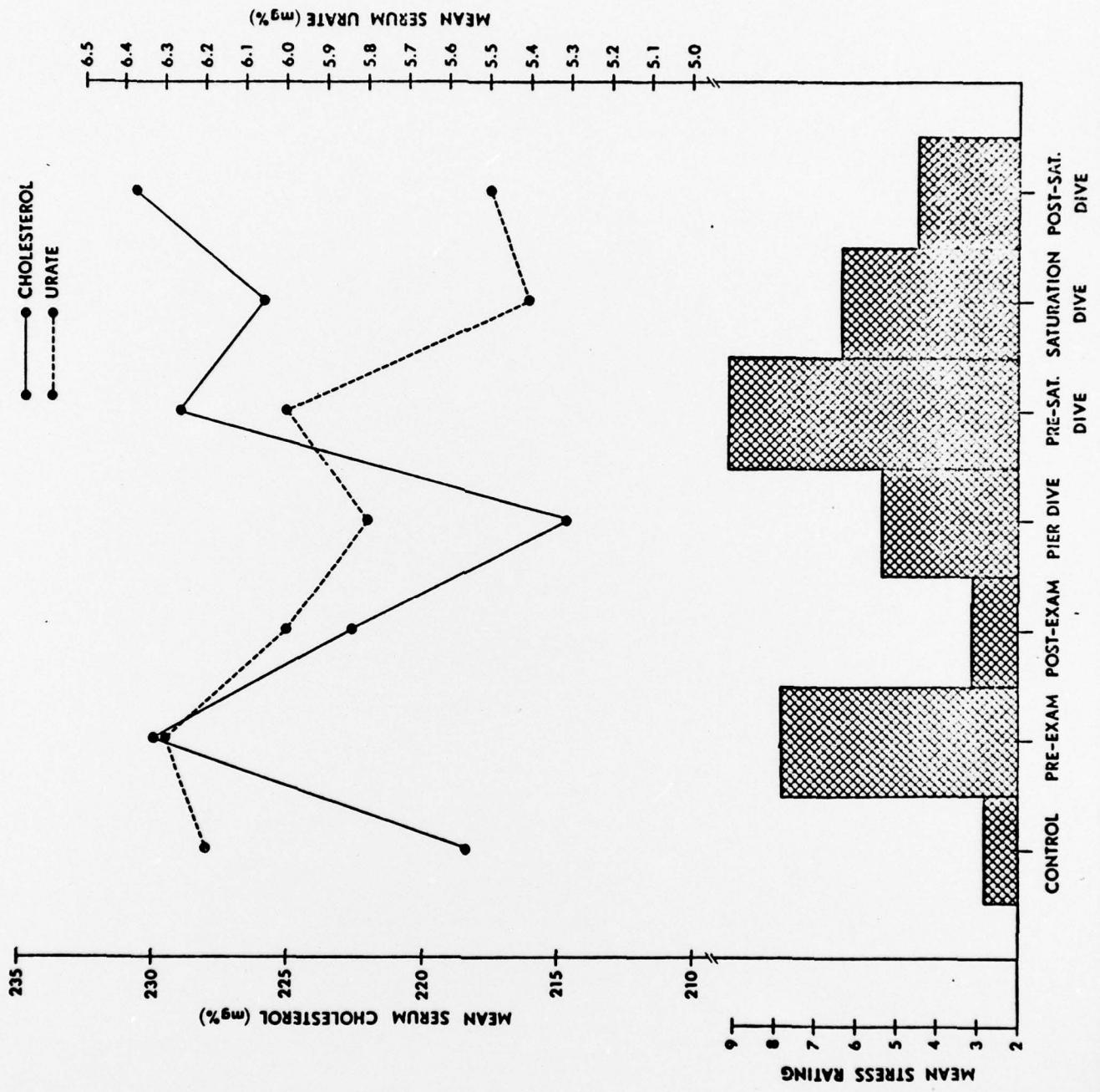
TABLE 1
Correlations of Diving Experience Items With Serum Cholesterol
and Uric Acid Levels

<u>Diving Experience Item</u>	<u>Prior to Exam</u>	<u>Prior to Saturation Dive</u>	<u>During Saturation Dive</u>
r with serum cholesterol			
Age (years)	.261	.414*	.491**
Education (years)	.103	.000	-.001
Years diving	.247	.401*	.466*
Years Diving. Age [†]	.085	.158	.203
Number of dives between 200 and 300 feet	.250	.378*	.523**
Number of dives [†] between 200 and 300 feet. Age	.213	.337	.503**
Number of dives made with hard- hat gear and HeO ₂ mixtures	-.385	-.250	.074
Number of dives made [†] with hard- hat gear and HeO ₂ mixtures. Age	0.374	-.182	-.229
r with serum uric acid levels			
Age (years)	-.110	-.249	-.129
Exotic diving experience (years)	-.343	-.443*	-.438*
Exotic diving experience. Age (years)	-.335	-.431*	-.439*

* p ≤ .05

** p ≤ .01

† partial correlation



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20. ABSTRACT (Continue on reverse side if necessary and identify by block number) Thirty-one U.S. Navy First Class Divers were studied over seven time periods encompassing varying levels of stress. All men were enrolled in a 14-week saturation diving training course. A 40-item mood questionnaire was administered and blood samples taken at each time period. Plasma levels for cholesterol and uric acid were determined. A Diving Experience Questionnaire was administered at the beginning of the course. Serum cholesterol was found		

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20. ABSTRACT (continued)

to rise to peak values during a classroom examination, decline during a pier dive, then rise and remain elevated throughout a saturation dive and ensuing decompression. Uric acid levels began high, showing a significant decline during decompression. Relationships between the two biochemical measures and the psychological parameters studied are presented.



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